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INJURY BY DISINFECTANTS TO SEEDS AND ROOTS IN SANDY SOILS.

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INTRODUCTION.

For several seasons the writer has conducted experiments in the application of disinfectants to pine seed beds for the purpose of controlling damping-off. Formaldehyde and various inorganic acids and salts have been tested. The work conducted at two of the nurseries with seed beds sown in the spring and summer has now been completed. The practical results of the disease-control work have already been briefly summarized.[1] Because of the interest of soil investigators as well as plant pathologists in the behavior of disinfecting agents in the soil, the data on injury to pine and weed seedlings by disinfectants are here published separately. Data on the effects of the disinfectants on the growth rate of pine seedlings are still being gathered from three nurseries, and it is hoped to publish these later.

Acknowledgments are due Dr. F. K. Cameron and others, of the Bureau of Soils, and Drs. Rodney H. True and F. D. Heald, of the Bureau of Plant Industry, for helpful suggestions.

SOIL CHARACTERS.

The nursery where most of the work was done is at Halsey, Nebr., in a valley among sand hills. The soil throughout the nursery area is quite uniform, both soil and subsoil being classed as fine sand. There is a fair amount of humus in the upper 10 to 12 inches, in some places extending to nearly 20 inches below the surface. Below 12 inches there is no humus in most of the nursery. The soil at the other nursery, that of the Pennsylvania Railroad, near Morrisville, Pa., is a light-gray sandy loam, with a fine, reddish, sandy subsoil which is rather nearer the surface than the subsoil at Halsey. Exami-

[1] Hartley, Carl, and Merrill, T. C. Preliminary tests of disinfectants in controlling damping-off in various nursery soils. *In* Phytopathology, v. 4, no. 2, p. 89-92, 1914.

nation by the Bureau of Soils of the United States Department of Agriculture shows the presence of the usual soil-forming minerals. The chemical and mechanical analyses are given in Table I.

TABLE I.—*Chemical and mechanical analyses of the nursery soils at Halsey, Nebr., and Morrisville, Pa.*

[The soil samples were taken from the upper 6 inches; subsoil from depths of 15 inches at Halsey and 12 inches at Morrisville.]

Analyses.	Percentage of soil.		Percentage of subsoil.	
	Halsey.	Morrisville.	Halsey.	Morrisville.
Chemical constituents:				
MnO	0.24	0.21	0.18	0.19
Fe_2O_3	3.08	1.60	2.85	1.30
Al_2O_3	14.93	8.72	14.95	6.20
K_2O	4.48	1.68	.80	1.88
P_2O_5	Trace.	Trace.	.48	.11
CaO	.86	2.23	3.79	1.35
Total salts by bridge	.21	.39	.09	.08
N	.07	.03	.09	.05
CO_2 (from carbonates)	None.	None.	None.	None.
Ignition loss (two determinations averaged)	2.41	2.93	.55	1.96
Lime requirements (CaO) per acre ... pounds	2,450	1,750	2,450	1,750
Mechanical constituents (size of particles):				
Fine gravel, 2 to 1 mm	0	1.0	0	0.5
Coarse sand, 1 to 0.5 mm	3.0	10.9	3.5	8.6
Medium sand, 0.5 to 0.25 mm	9.5	16.1	15.4	13.4
Fine sand, 0.25 to 0.1 mm	58.1	28.9	61.3	30.5
Very fine sand, 0.1 to 0.05 mm	21.0	19.2	17.5	23.5
Silt, 0.05 to 0.005 mm	6.5	18.5	1.5	18.1
Clay, 0.005 mm. and finer	2.1	5.5	.8	5.2

The wilting coefficient, determined by the indirect method of Briggs and Shantz,[1] was 3.42 per cent for the surface soil and 1.5 per cent for the subsoil at Halsey, and 4.92 per cent for the surface soil and 4.73 per cent for the subsoil at Morrisville. The samples examined from Halsey were taken from 10 different points in the nursery, while the samples from Morrisville represent three different points.

EXPERIMENTS AT HALSEY, NEBR.

Experiments at the nursery at Halsey have been carried on in cooperation with the United States Forest Service during the past five years. Mr. Robert D. Rands assisted the writer during the year in which most of the data were secured, and Messrs. R. G. Pierce and Fred R. Johnson, of the Forest Service, rendered material assistance in the work.

DISINFECTANTS USED.

Part of the sulphuric acid used in the following experiments was C. P. (chemically pure), but most of it was a clear commercial grade, the acid used in most of the work here reported having a specific gravity of 1.84 and that used for the latest work a specific gravity of

[1] Briggs, L. J., and Shantz, H. L. The wilting coefficient for different plants and its indirect determination. U. S. Department of Agriculture, Bureau of Plant Industry Bulletin 230, 1912.

1.83. Repeated parallel tests of C. P. and commercial sulphuric acid failed to develop any difference in their effect on the seed beds. A part of the hydrochloric and nitric acids used was C. P. and part commercial. The ammonia used was the strongest commercial ammonia water obtainable from local druggists (ordinarily 26° Beaumé). The formaldehyde used was the so-called 40 per cent commercial solution. Because of the need of distinguishing between pure formaldehyde and this commercial solution the latter will be referred to as formalin. The general use of the term "formalin" for the commercial solution appears to have become approved by custom,[1] despite the fact that this term formerly applied only to the product of an English firm. The lime-sulphur used was a commercial solution with a specific gravity of 1.31. The mercuric chlorid used was C. P. and the cupric sulphate was the fully hydrated crystalline form. The copper acetate was neutral, containing a single molecule of crystallization water. The zinc chlorid was a technical grade, granular, guaranteed from 95 to 98 per cent pure. All lime used was air-slaked.

The unit of measure used throughout is the fluid ounce (29.574 c. c.) for the acids, formalin, ammonia, and lime-sulphur solution, and the avoirdupois ounce (28.35 grams) for the other substances. Except where otherwise stated, all of the disinfectants were applied in aqueous solution. When lime was used the powder was spread dry on the surface of the bed and was worked into the upper 2 or 3 inches with a rake. Two or three pints of water per square foot of seed bed was found a convenient vehicle for applying the disinfectants. Because of the variable moisture content of the soil the degree of dilution of the solution before application is not of the greatest significance. The amount of the disinfectant used per square foot of soil surface is given in all cases as the measure of the strength of the treatment.

PLANTS UPON WHICH OBSERVATIONS WERE MADE.

The seed beds on which disinfectants were used were sown with different species of pine. Jack pine (*Pinus divaricata*) was the species used in most of the work, while western yellow pine (*P. ponderosa*), Norway pine (*P. resinosa*), and Corsican pine (*P. laricio*) were also used, the relative frequency being in the order named.

Weeds of various types appeared in the seed beds in addition to the pines, and data as to their tolerance of disinfectants were also obtained. Cryptogams were represented by a large-stalked species of Equisetum, the algæ conspicuous in many nurseries being present to but a slight extent. Monocotyledons were represented by various grasses, *Eragrostis cilianensis*[2] being much the most common, while *Echinochloa crus-galli*, *Panicum barbipulvinatum*,[2] and *Chaetochloa*

[1] Perkin, W. H., and Kipping, F. S. Organic chemistry, new ed., p. 124. London, 1911. See also Webster's New International Dictionary, 1913.

[2] Determinations made by Mr. P. L. Ricker.

viridis[1] were also more or less common. The nurserymen pulled up most of the weeds before flowering, so that it was not possible to determine positively the relative frequency of the different grass species for each plat. The commonest dicotyledons were *Mollugo verticillata*,[1] *Portulaca oleracea*,[1] *Amaranthus retroflexus*,[1] *A. hybridus*,[1] *A. graecizans*,[1] *A. blitoides*,[1] and *Euphorbia glyptosperma*.[1]

INJURY TO PINES BY SULPHURIC ACID APPLIED AT OR AFTER GERMINATION.

In the following cases sulphuric acid was applied to the beds after some pine seedlings had come up. Because of the great irregularity of germination in many beds the time of germination can be given only approximately. It represents as far as possible the date by which enough seedlings had appeared to constitute a fair stand. Most of the experimental plats were sown with jack pine. The results with this species appear in Table II.

TABLE II.—*Effect of sulphuric acid on seedlings of jack pine, at Halsey, Nebr.*

Number of plats treated.	Time of treatment.	Fluid ounce of acid per square foot.	Volumes of water.	Result.
4	On date of germination	0.172	128	All killed.
	6 days after germination	.086		
2	On date of germination	.086	256	Nearly all killed.
	6 days after germination	.043		
	8 days after germination	.043		
	13 days after germination	.043		
4	1 day after germination	.086	128	Many killed.
4	....do	.043	256	More killed than in preceding experiment.
	3 days after germination	.043		
	6 days after germination	.086		
	8 days after germination	.043		
	21 days after germination	.043		
2	On date of germination	.021	512	Germination, 11.8 per cent.
	2 days after germination	.021		
2	On date of germination	.011	1,024	Germination, 13.8 per cent.
	2 days after germination	.011		
2		None.		Germination, 14.7 per cent.

Half of the plats in Table II which were given the stronger solutions were sprinkled lightly with water immediately after each treatment. This watering had no evident effect in the plats treated with the 128-volume solution, but in four plats which received the 256-volume solution, followed by sprinkling, the stand of seedlings was more than twice as great as on four adjacent plats which were given the acid treatment only.

The results in the plats treated with the 512-volume solution indicate that a total of 0.043 ounce of acid per square foot applied before germination was complete was sufficient to prevent the appearance of some of the latest germinating seedlings, while 0.021 ounce in two applications had little or no effect. Further tests would be necessary to prove that injury can be caused by these very weak treatments.

[1] Determinations made by Mr. P. L. Ricker.

Acid was also used after germination on seed beds of western yellow pine. In the first test the percentage of the seedlings which died during the first 33 days after germination was determined for four plats, as follows:

Plat VIII-A.—On the twelfth day after germination, 0.086 ounce of acid in 128 volumes of water; repeated on the fourteenth and nineteenth days. Loss, 72 per cent.

Plat VIII.—Same acid treatment as VIII-A, but sprinkled lightly with water after each application. Loss, 33 per cent.

Plat 27.—On the sixth and sixteenth days after germination, 0.086 ounces of acid; 12, 14, and 19 days after germination, 0.043 ounce of acid; solution in 256 volumes of water. Loss, 21 per cent.

Plat 28.—No treatment. Loss, 23 per cent.

While the loss in plat 27 was slightly less than that in the untreated plat there is clear evidence that the acid killed the seedlings, as the parasitic loss in this plat was very much less than in the untreated plat.

The treatments on Plats VIII and VIII-A were practically duplicated on a seed bed 13 days younger, with the result that the losses for the first 20 days were 45 and 47 per cent, respectively, as compared with 16 per cent in the nearest check.

Further tests of sulphuric acid on germinating yellow pine were made during the two following seasons. In the first case, acid in 256 volumes of water was tested on beds which had received 0.188 ounce of formalin per square foot 40 days before sowing, a treatment which in itself had no appreciable influence. The results were as follows:

Plat 402-S.—Seven and again twenty-five days after germination, 0.125 ounce of acid. Germination, 64 per cent; loss after germination, 44 per cent.

Plat 402-N.—Seven days after germination, 0.125 ounce of acid. Germination, 51 per cent; loss, 30 per cent.

Check plat.—No acid. Germination, 68 per cent; loss, 62 per cent.

In this series, the effect of the acid was clearly to prevent the appearance of the latest germinating seedlings and to kill the youngest seedlings which had already broken through the soil. The heavier loss in the untreated plats is due to heavy parasitism, which the acid treatment almost entirely prevented.

The following season, using a solution of one part in 256 volumes of water, the following amounts of acid were applied to yellow-pine plats: 0.047 ounce per square foot on two plats three days after germination; the same amount on two other plats six days after germination; and 0.063 ounce on three plats seven days after germination. No noticeable injury occurred, though counts of the seedlings indicate that a few were probably killed by the acid.

Most or all of the injury caused by applications after the beginning of germination was due to injury to the roots. The light sprinkling with water just after acid applications, which in a number of

cases resulted in lessening injury, presumably exerted its effect through an immediate further dilution of the acid in the surface layer of soil. While part of the apparent freedom of the aerial parts of the plants from direct acid injury may be due to the slight tendency of liquids to adhere to pine seedlings, drops of 1 to 256 acid solution by volume (0.71 per cent by weight) frequently remained caught in the center of the whorls of cotyledons of yellow-pine seedlings. This localization of solution was not accompanied by any noticeable localized injury. The experience of Craig,[1] indicating direct injury to the foliage of grapes, plums, and apples out of doors by a solution containing but 0.25 per cent of the acid, was more closely duplicated in the case of seedlings of a grass resembling a common native species of Panicum, which occurred in some of the plats. Definite characteristic spots of dead leaf tissue were noted on the grass plants in a few cases in plats treated with a solution of 1 to 512 by volume (0.36 per cent). The solution adhering to the leaves is, of course, concentrated by evaporation of the water after application, so the injury from spraying with solutions is actually caused by a much stronger solution than that applied.

The tests outlined in the foregoing statement indicate that after the seed begins to germinate, any application of sulphuric acid sufficient to affect materially the activity of the damping-off parasites will cause the death of the radicles of some of the pine seedlings.

In applications after the beginning of germination, the concentration of the solution applied, as well as the amount of acid used per square foot, seemed distinctly related to the amount of injury to the roots of the seedlings. This indicates that the injury occurred very promptly after the application of the solution, before diffusion between the upper and lower layers of soil had time to equalize quantities and concentration of the soil solution. The younger parts of the roots were still in the upper 1 or 2 inches of soil in most cases at the time the injurious solutions were applied.

INJURY TO PINES BY SULPHURIC ACID APPLIED AT THE TIME OF SOWING.

In applications made at the time of sowing it was found that stronger treatments could be given without injury to the pines than when the treatments were delayed until germination. Stronger treatments were also required in order to control parasitic fungi, so that it was necessary in these tests also to work with treatments strong enough to cause injury to seedlings. Because of the numerous advantages of acid treatment at sowing, from the standpoint of disease prevention and nursery practice, a detailed study of the injury it causes to seedlings was undertaken with a view to prevention.

[1] Craig, John. Effects of dilute sulphuric acid on foliage. *In* Canada Exp. Farms, Rpt., 1893, p. 101-102, 1894.

The procedure followed in treatment at sowing time was to (1) prepare the seed bed, (2) soak it with the disinfectant, (3) sow the seed broadcast, (4) cover with one-fourth inch of dry soil, and (5) apply the rest of the solution. The seed bed was not stirred up after the application of the solution was commenced. In no case in spring-sown beds has there been any indication that the treatments injured the pine seed before germination started, although the treatment, in strengths varying from 0.125 to 0.375 fluid ounce of acid per square foot, has been tested during the past three seasons in 19 different experimental series of jack pine, in 4 series each of yellow pine and Norway pine, and 1 series of Corsican pine. The proportion of germination in acid plats was nearly always higher than in the untreated plats (due to the prevention of parasites rather than to stimulation), and as high as in plats of soil disinfected by heat.

In jack-pine plats in which germination was reasonably prompt (12 to 14 days) and no special measures were taken to prevent injury to seedlings, many seedlings were killed or injured after germination began on plats which had received, respectively, 0.125 ounce and 0.141 ounce of acid per square foot at sowing, while 0.188 ounce per square foot always resulted in injury unless special protective measures were taken.

DESCRIPTION OF THE INJURY.

Injury to the seedlings in plats treated at or before the time of sowing took the form of damage to the growing apices of the radicles, with the result that extension of the root was stopped. Whether the meristematic apical cells were actually killed or simply lost their meristem qualities was not determined, though the former is the more probable. In most cases, root apices rendered incapable of growth retained their normal cream color for a few days after the injury and often recovered, though in severe cases they turned dark very soon. Plate I and text figures 1 and 2 show chemically injured seedlings. Plate I, figure 1, shows a healthy seedling, younger than the injured seedlings in figures 2, 3, and 4 of this plate, so that the darker color of the upper parts of the roots of injured seedlings is chiefly due to difference in age, rather than to the effects of the acid. The disproportionately short roots of the injured seedlings are especially noteworthy.

FIG. 1.—*Pinus divaricata* injured by acid. Root growth has just been resumed by two laterals after 11 days suspension. × 1⅓.

Ordinarily the growth of cells just back of the apex was not entirely prevented, so that the root tips became truncated as a result of the uneven growth. (Pl. I, fig. 2.) Distorted growth was also common. The capacity for absorption was usually retained by the injured roots for some

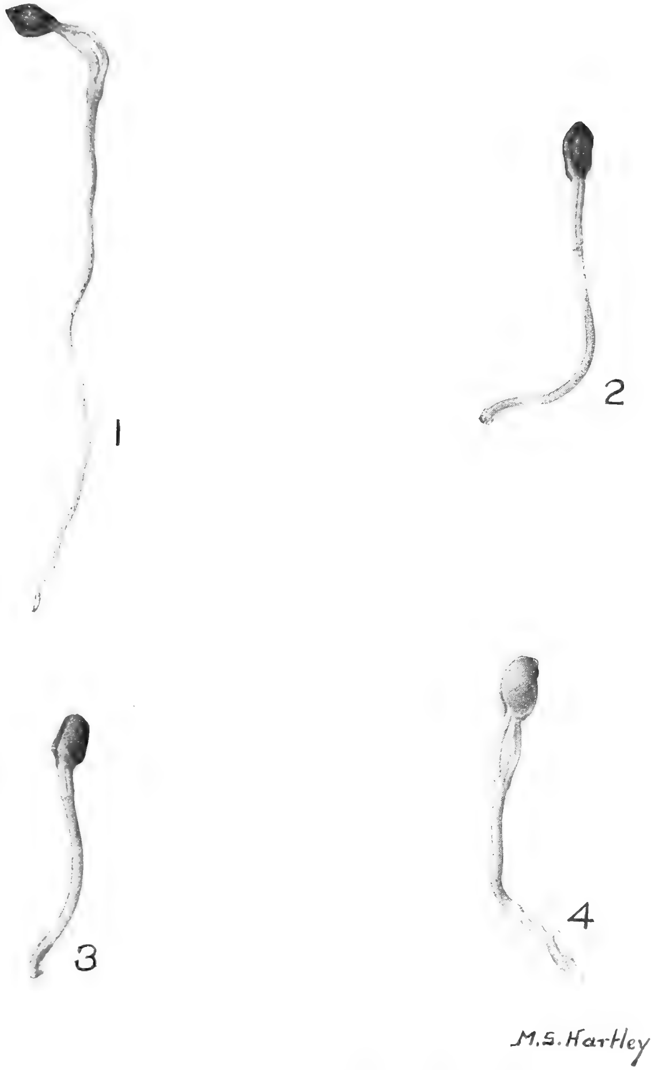
time. Although injury to root apices commonly took place before the seedlings appeared above the soil, most injured seedlings came up, and when the soil around the short root was kept moist the growth of the stem and leaves continued for some time at a normal rate. All of the development of the aerial parts of the seedlings shown in Plate I, figures 2, 3, and 4, was made after the extension of the root had been stopped by acid.

Injured seedlings ordinarily lived till the surface of the upper part of the root became brown and presumably impervious, as in the older parts of the root in healthy seedlings after two or three weeks. In the worst injured seedlings this root browning seemed to take place somewhat earlier than in healthy plants. The decrease in diameter which is noticed in the older parts of normal roots at the time of browning was seldom observed in acid-injured roots. Because the injured seedlings were not able to develop new root tissue, absorption ultimately became impossible and death from drought ensued. The seedlings shown in Plate I, figures 2 and 3, have practically reached this condition, though both still appeared to be growing normally when they were dug up. Plate I, figure 4, shows a seedling injured at the same time as that in Plate I, figure 3, which has recovered by recommencing root growth.

FIG. 2.—*Pinus ponderosa* injured by copper sulphate. Root growth has been resumed by a number of laterals. (Natural size.)

Where the roots of injured seedlings were very short, the plants died very soon, either because the soil was allowed to dry out to below the level reached by the short root or because the short root did not afford sufficient mechanical support for the top-heavy stem, and the seedling fell over or was washed out in watering. In the cases where injury was earliest, so that the radicle had scarcely emerged from the seed coat by the time its tip was killed, the seedlings failed to appear above ground at all.

In a good many cases seedlings which had extended their roots a centimeter or more before injury ultimately recovered, either because of a resumption of terminal root growth, as shown in Plate I, figure 4, or by laterals starting just back of the apex, as in text figure 1. In such cases the parts of the seedlings above ground at no time showed any effect of the acid, and the only way in which the existence of injury could be detected was by examining the roots. Renewal



HEALTHY AND ACID-INJURED PINE SEEDLINGS.

FIG. 1.—*Pinus divaricata*, healthy seedling. (×2.) FIG. 2.—*P. divaricata*, acid injured. (×2.) Probably not capable of recovery. The root growth was stopped before the seedling came up. The entire development of the stem and leaves above ground has taken place since the cessation of root growth. FIG. 3.—*P. laricio*, acid injured. (×2.) Injured when so little root had developed that there was no possibility of a resumption of growth. Illustration made 10 days after the killing concentration occurred. FIG. 4.—*P. laricio*, acid injured. (×1½.) Recovering by terminal resumption of root growth, as shown by the white root tip.

of root growth in injured seedlings was most commonly observed from 8 to 12 days after the original cessation of growth. Dr. Perley Spaulding has advised the writer that a year prior to the observations here reported he found this resumption of growth by laterals in injured western yellow-pine seedlings in experimental plats at Burlington, Vt.

It is seldom possible to recognize acid injury immediately after occurrence. Even after death takes place it is not possible to distinguish the deeper rooted injured seedlings from those killed by parasites, as by the time the seedling gives indications of death above ground the roots are too badly decayed to show what caused death. The best way to detect acid injury is to dig up healthy-looking seedlings in different parts of a plat a week or ten days after the first seedlings come up. The roots of the seedlings will be found to have the following characters:

(1) Acid-injured seedlings (Pl. I, figs. 2 and 3). Length, one-fourth to five-eighths of an inch. Color, if brown at all, tip will be as brown as the rest; root firm throughout.

(2) Healthy seedlings (Pl. I, fig. 1). Length, 1 to 3 inches. Color, upper part may be brown, but tip will be white.

(3) Damped-off seedlings (attacked by parasites). Length, usually same as healthy, but lower part may be entirely decayed, making root appear short. Some part of root examined will always be found soft from decay, while acid-injured roots are firm throughout.

NOTE.—Care is needed to distinguish between the short root of an injured seedling and a healthy root which has been broken off short by accident. With a little practice, the difference between a root tip and a broken end can be easily recognized.

PREVENTION OF INJURY BY LEACHING.

The first attempt to prevent injury to germinating seedlings from the residue of acid applied at sowing was by leaching. To different plats in a bed which had received 0.188 ounces of acid at sowing three days earlier, 4, 8, 12, and 16 pints of water per square foot, respectively, were applied. The plats were thereafter given sprinklings equal to 0.3 of an inch of rain often enough to insure germination, which took place 11 days after sowing. The heaviest initial watering, equivalent to 3.2 inches of retained rainfall, prevented most of the injury which occurred on the other plats, but not all. The plat receiving but 4 pints (0.8 inch) suffered heavily, while the amount of injury in the 8 and 12 pint plats was intermediate. In a second test, with an acid treatment of 0.211 ounce at sowing, followed by germination in eight days, a 6-inch watering was given three days after sowing. The bed was purposely allowed to become quite dry on the day of germination, and later examination showed that a small number of the pines were injured. In a third test, this 6-inch watering was used on a bed which had received 0.313 ounce of acid. The bed was allowed to become somewhat dry 10 days

after the acid treatment (1 day before germination), and a number of seedlings were injured. It was evident from the results obtained that these heavy applications of water leached out enough acid materially to reduce acid injury. Leaching is evidently not practicable as a method of preventing injury at most nurseries when germination is prompt. In a sandy soil when the weather is cold and germination requires 18 or 20 days, leaching soon after the application of acid may be a practicable method of preventing injury.

PREVENTION OF INJURY BY FREQUENT WATERING.

Fortunately two definite relationships which opened the way for developing a practicable method of controlling the injury to the pines were found. It was found that the amount of water in the soil at the time of germination bore a direct relation to the amount of injury, and that injury seldom occurred after the seedlings had sent their roots down five-eighths of an inch into the soil. The length of root shown in Plate I, figure 3, is typical of injured seedlings in general. The stoppage of growth of root apices in treated beds always occurred at times when the upper soil became relatively dry and while the root tips of germinating seedlings were still in the upper five-eighths inch of soil. Although the nurserymen water the beds often enough to prevent drought injury to the seedlings, great variation in the moisture content of the surface soil occurs. The upper one-fourth inch of soil at this nursery just after watering has frequently been found to contain 21 to 25 per cent of moisture, while at the same points the soil when dry has contained but 1.96 per cent of water, the average of 12 determinations made on different occasions. In a single period of 11 hours the moisture content of the surface soil at four different points in the seed beds dropped from 12.02 to 1.85 per cent. This of necessity caused great variations in the concentration of the soil solution. While beds were not ordinarily allowed to become as dry as this during the germinating period, they often became quite dry at the surface. A little below the surface the moisture content of the soil is more stable. The most rapid loss of moisture found in the seed beds from 1 to 2 inches in depth during the period in which determinations were made was a drop from 17 to 11½ per cent in a period of approximately 36 hours. This explains the relative safety of roots which have penetrated below the upper half inch of soil. That the root above the tip should resist relatively high concentrations of acid is in agreement with the results of Heald [1] and other investigators, who find the tip of the root to be the portion most sensitive to poisons. The difference in resistance between the very

[1] Heald, F. D. On the toxic effect of dilute solutions of acids and salts upon plants. *In* Bot. Gaz., v. 22, no. 2, p. 130, 1896.

tip of the root and the tissue just back of it is well shown by the location of the new laterals developed by the seedling in figure 1.

In addition to the increased concentration of the acid solution already in the surface soil, due to the decrease of the solvent, acid from lower levels is presumably brought up to the surface by the capillary rise of the soil solution to replace that lost by evaporation. When the treated soil is soaked thoroughly with water and subjected to continuous evaporation for several days, but at a rate slow enough to avoid drying the surface soil entirely and breaking the capillary connection, this continuous upward movement of solution ultimately results in killing concentrations in the surface soil, even while it is still very moist. The problem of preventing injury to seedlings therefore becomes one of not only keeping the surface soil moist, but of maintaining a fairly constant downward movement of soil moisture, or at least of preventing a continuous upward movement for any considerable period, until after the roots of all seedlings have extended half an inch into the soil. Experience has shown that this can be done more easily with frequent light waterings than with heavier and less frequent applications.

A very few hours' delay in watering at a critical time has in some cases been enough to cause the killing of root tips by acid. Under certain conditions, as outlined in the foregoing paragraph, injury occurred before the beds appeared at all dry at the surface. Since appearances could not be relied on to show when watering was needed, systematic watering was tested. Furthermore, variation in individual judgment made necessary the use of measured quantities of water. Daily waterings equivalent to 0.4 of an inch of rain were not in all cases sufficient to prevent injury entirely. However, half this quantity applied twice as often, with the soil wet to begin with, was found sufficient to prevent all injury from moderate amounts of acid, even in very hot, dry weather. For large beds at this nursery which have received 0.188 ounce of acid at sowing, watering equivalent to 0.3 of an inch twice daily during the germination period has been recommended for summer use, so as to make certain that in the necessarily uneven large-scale work all parts of the bed will get at least 0.2 of an inch at each watering.

For work in cold spring weather, when the germination period is long, the expense of this special watering becomes considerable, and it further cools the soil to such a point that germination may be still more delayed. No such frequent watering is necessary to prevent injury in cool weather, but because of occasional hot, dry weather in early spring it is not safe entirely to abandon watering twice daily. A rather extreme instance of the variable temperature at Halsey was the rise of the temperature, as shown by a Weather

Bureau thermometer under a shelter 4 feet from the ground, from 37° F. at 8 a. m. to 98° F. at noon of the same day in April. The evaporation from white porous-cup atmometers set in the seed beds has varied from 14 to 59 c. c. for 24-hour periods 10 days apart, and still greater variations are to be expected from the darker soil surface. Hot, dry days increase the danger from acid injury both by increasing water loss and consequent acid concentration and by hurrying germination before the acid solution in the upper soil has had much time to decrease in strength. In view of the variability of weather conditions, the system now followed in preventing acid injury is to water daily in ordinary spring weather, every other day or even less often in misty or rainy weather, and twice daily when the temperature exceeds 80° F. In clear weather, waterings are to approximate 0.3 of an inch, while in cold and cloudy weather 0.2 of an inch is to be used. This watering system has proved practicable, and has been entirely successful in preventing injury to pines from acid applied at the time of sowing.

RELATION OF STRENGTH OF TREATMENT TO EXTENT OF INJURY.

The degree of dilution of the sulphuric acid in applications at sowing had no apparent relation to the amount of injury likely to result to the seedlings; that is, if 0.25 ounce of acid per square foot was applied, it made no difference, so far as noticed, whether it was dissolved in 64 or 192 volumes of water. There was not a sufficient number of tests with this factor as an independent variable to establish an entire lack of relation, but it is quite certain that within the limits given the amount of water used in making up the solution is not an important variable.

The first results indicated a rather surprising lack of constant relation between the amount of acid used per unit of soil surface and the amount of injury. In an early test of varying amounts of acid, all of which caused considerable losses of seedlings, the final stands in the plats were as follows:

Series 501.—Jack-pine plats; all except the check plats were treated with acid at sowing.

Eight check plats untreated. Final stands ranged from 71 to 163 per square foot; average, 122.

One plat, 0.125 fluid ounce of acid per square foot at sowing. Final stand, 216.

One plat, 0.141 ounce of acid. Final stand, 118.

Two plats, 0.188 ounce of acid. Final stands, 191 and 143; average, 167.

Three plats, 0.234 ounce of acid. Final stands, 107, 110, and 80; average, 99.

Two plats, 0.250 ounce of acid. Final stands, 23 and 153; average, 88.

One plat, 0.313 ounce of acid. Final stand, 94.

Two plats, 0.375 ounce of acid. Final stands, 11 and 116; average, 64.

In this series, as in those reported in the remainder of this paper, the plats received weights of seed proportional to their area.

In this series the variation between individual plats is great. Especially in the cases of the 0.250-ounce and the 0.375-ounce plats the variation between plats given the same acid treatments is much greater than the average variation between plats given different treatments or between the untreated plats, which are subject to much heavier variation from the action of parasites than the acid-treated plats. However, the averages indicate a distinct increase in the amount of injury as the quantity of acid is increased. The great individual variation between plats with the same acid treatment is to be explained by two factors which were not controlled. In the first place, different plats germinated at somewhat different times. Some plats therefore had a much greater average root length than others at the time the killing concentrations of the soil solution occurred. This greater root length resulted in the sensitive tip being farther down in the soil, where the acid solution does not become as concentrated as in the soil at the surface. It may also have been true here, as found by McCool[1] in his work with barium, strontium, sodium, and ammonium, that the root tips of seedlings a few days old are less susceptible to injury than those of seedlings which have just germinated, so that the age of the seedlings may have been even more important than the location of the root tips in making the older seedlings more resistant. Furthermore, those with the longer roots were not only less likely to be injured but also had a better chance to recover. (Compare Pl. I, figs. 3 and 4.) A more important variable factor in causing different results in plats with identical acid treatments was the watering during the germinating period. While all plats were watered at the same time, no attempt was made in series 501 to secure special uniformity in watering, and some became drier than others. A later test of different amounts of acid was made with plats sprinkled with measured quantities of water twice daily during the germination period. Germination took place nine days after the plats were treated and sown. The results are given in Table III.

TABLE III.—*Relation of the amount of acid applied and the thoroughness of subsequent waterings to the death of pine seedlings on plats treated with sulphuric acid at the time of sowing.*

[Seedlings per square foot surviving 44 days after germination.]

Water per square foot.	Treatment (ounces of acid per square foot).		
	0.211	0.250	0.313
2 pints at each watering........seedlings..	....	179	142
1.5 pints at each watering........do....	281	151	91
1 pint at each watering........do....	125	64	47

[1] McCool, M. M. The action of certain nutrient and nonnutrient bases on plant growth. N. Y. Cornell Agr. Exp. Sta. Mem. 2, p. 159-162, 1913.

The decrease in stand both with decreasing amounts of watering and with increasing amounts of acid was sufficiently consistent in this experiment to establish beyond a reasonable doubt the relationship, both of the amount of acid used and of the amount of watering done, to the acid injury. In the weakest acid plat with the intermediate watering, no appreciable injury occurred. Because of the variation in germination aside from the influence of acid, the results were not always quite as consistent as in this series, but no reason has been found to doubt the relation between the amount of acid and the extent of injury in beds treated at sowing.

INJURY TO PINES BY SULPHURIC ACID APPLIED BEFORE SOWING.

In treating beds with sulphuric acid to kill fungous parasites the attempt was made to evade toxic action on the seedlings by applying the acid a number of days before sowing. Jack pine was also used in most of these tests. In such cases the beds were ordinarily hoed and raked just before they were sown, so that the upper 2 or 3 inches of soil was well mixed after the acid was applied. In the plats treated at sowing there was the possibility that the injury was limited to the surface five-eighths of an inch of soil, simply because this layer of soil had acted as a trap for the acid, absorbing most of it at the time of application. In the case of plats treated before sowing there was no such possibility. The seeds were in most cases covered with about one-fourth of an inch of soil taken from the upper 1 to 1½ inches of the soil of a near-by area that had been given the same treatment as the plat sown. Considerable injury occurred in plats which received 0.25 and 0.375 ounce of acid nine days before sowing (20 days in all elapsing before germination), although the treated plats received approximately 1.6 inches of water five days after sowing, followed by 0.3 to 0.4 of an inch daily till after germination. The slight drying of the surface soil which resulted in the injury on these plats took place the first day after germination, 21 days after the application of the acid.

In another series, using the same species of pine, amounts of 0.281, 0.375, and 0.687 ounce of acid per square foot were applied 11 days before sowing, two plats receiving the latter amount. Four days after sowing, the plats were given approximately 1.6 inches of water, followed by waterings of approximately 0.3 to 0.4 inch on the sixth, eighth, ninth, tenth, and eleventh days from sowing. Germination took place on the eleventh day, 22 days after the application of the acid, and on the morning of this day the soil surface became somewhat dry, but not dry enough to cause appreciable drought injury in the nonacid plats. As shown by later examination of the length of the acid-injured roots, injury took

place at this time. It was most serious in the 0.375-ounce plat, mainly because it had become somewhat drier than the rest. Even the 0.281-ounce plat seemed more injured than the 0.687-ounce plats, which were not seriously affected. The activity of parasites, mostly, probably, *Pythium debaryanum*, in the soil in these plats during and after the time that this injury was occurring to the seedlings is a matter of some interest. The slight relationship between the amount of acid used and the amount of injury taking place in these plats 22 days after treatment emphasizes what has already been said as to the apparent equalization of strength of acid solutions of different original strengths in the soil as the concentration decreases.

Plats of jack pine which had been entirely killed by applications of 0.172 ounce of acid at the date of germination and 0.086 ounce six days later, 0.258 ounce in all, were resown with the same species 23 to 24 days after the first treatment, germination taking place 34 to 36 days after the first treatment. No serious injury occurred to the seedlings in this second sowing, though no special watering was given. Similar results were obtained with yellow pine in plats treated with 0.3 ounce of acid 39 days before sowing (50 days before germination), no serious injury occurring despite the entire lack of any special watering. In all cases, acid applied before sowing can be kept from causing injury quite easily by the watering methods used for beds treated at sowing. The tests indicate that if germination takes place at any time during the first month after 0.25 ounce of acid is applied to the beds it will be necessary to give more than the usual nursery watering during the germination period in order to insure freedom from injury to the seedlings. Though it is somewhat easier to prevent acid injury in beds treated several days before sowing, treatment at the time of sowing is so much more effective against the damping-off parasites that it is considered preferable.

RELATIVE RESISTANCE OF VARIOUS SPECIES OF PINE TO SULPHURIC ACID.

There was considerable difference in the amount of injury caused by similar acid treatments on different species of pine. Jack pine, as a rule, seemed most liable to serious injury, while yellow pine was least often damaged, and Norway and Corsican pines were intermediate. The resistance of yellow pine as compared with jack pine was especially evident in beds treated shortly after germination. Most of this apparent difference in resistance is due not to variations in the capacity of the root tips to endure acid, but to a difference in the rate of growth. Yellow pine has a seed approximately ten times as heavy as that of jack pine and sends its root down much

faster at the start. By the time a yellow-pine seedling breaks through the soil cover its root has gone down much farther into the soil than with jack pine at the same age, and the application of a disinfectant to the soil surface at this time is therefore much less likely to injure the yellow-pine root tip. When disinfectants are put on the soil at sowing, the root tips have not yet emerged from the seed, and yellow pine has no such distinct advantage over jack pine. There is still a difference in depth of planting, however, as yellow-pine seeds are usually covered deeper than those of jack pine and the root tips thus start at a lower level. The more rapid growth is also of some advantage in beds treated before germination, as injury occurs only at times of surface concentration. The root tips of yellow pine may get down far enough to avoid injury from a concentration which occurs before the tips of jack-pine roots have reached the safety zone. While yellow pine has been less often injured than jack pine by acid applied at the time of sowing, concentrations occurring while there was a large porportion of yellow-pine root tips in the surface soil have killed large numbers of seedlings. In one extreme case, in which 0.250 ounce of acid per square foot was applied 28 days before sowing and repeated at sowing, with germination following five to six days later, only two-thirds as many seedlings came up as in untreated plats, and of these over 90 per cent died, nearly all as a result of acid injury. On the whole, while yellow pine has been much less often injured by acid treatment, the evidence indicates little, if any, greater resistance of its root tips than that shown by jack pine.

Corsican pine shows injury in the same way as jack pine (Pl. I, figs. 2 and 3). It has a seed smaller than yellow pine, but still much larger than jack pine and producing a faster initial root growth. It therefore seems a little less liable to injury than jack pine, for the same reasons that yellow pine is less liable. Norway pine on the other hand, though having a larger seed than jack pine, makes a much slower initial root growth at this nursery. Its slightly longer germination period gives the acid more time for dissipation, but the indications are that the root tips of this species possess a slightly greater acid endurance than those of jack pine. Corsican and Norway pine have not been tested as much as the other two species, and the evidence obtained as to their relative resistance has less value.

INJURY TO MISCELLANEOUS PLANTS BY SULPHURIC ACID.

The watering given pine seed beds at the Halsey nursery resulted in the germination of great numbers of previously dormant weed seeds of the species listed on pages 3 and 4. These ordinarily began to appear a little later than the pines and continued to come up in con-

siderable quantities for the first two or three weeks, after which time the number which came up decreased.

Most of the data on the effects of sulphuric-acid treatments on weeds were obtained on beds treated at the time of sowing. The observations indicated marked differences between the species observed in their ability to grow in soil recently treated with acid. It was evident throughout that the pines were less easily injured than most of the weed species. On plats which received no special watering till after germination, 0.125 ounce and 0.141 ounce of sulphuric acid per square foot, respectively, at the time of seeding entirely prevented weed growth. The untreated plats in this series were fairly well covered with Portulaca and grass species and with a few plants of Amaranthus. At sowing in another series on a plat given very frequent watering, 0.125 ounce of acid failed to reduce perceptibly the number of common weeds. Another plat given the same treatment, which had also received 0.125 ounce of acid 13 days before sowing, showed entire freedom from weeds, with only partial injury to the pines. In repeated tests during successive seasons, treatments of 0.188 ounce of acid at the time of sowing regularly prevented practically all weed growth for the first three weeks after the germination of the pines. In some cases no weeds came up in treated beds until a month after the appearance of the pines. Beds treated with acid and so watered as entirely to prevent injury to the pines were nevertheless so free from weeds as a result of acid application that the cost of weeding the treated beds during the whole season has been only one-third that of untreated beds.

The appearance of Equisetum in acid-treated plats was of some interest. In an insufficiently watered acid plat on which the pines were seriously injured and on which not a single phanerogamic weed appeared, more Equisetum developed than in most of the untreated beds in the nursery. Equisetum was not a common weed anywhere, but it occurred more frequently in the acid beds than in the beds not treated.

The grasses throughout gave evidence of greater ability to endure the acid applied to the soil than did the dicotyledons. They were usually the predominant weeds and often the only ones in acid plats. This greater predominance of grasses over dicotyledons in the acid plats left little doubt as to their superior endurance of this treatment.

Unfortunately, few data were secured as to the factors which controlled the varying capacity of the different plants observed to endure acid applied to the soil. Most of the injury to the weeds did not occur in just the same way as to the pines. In the pines the commonest phenomenon was root injury, which allowed the seedlings

to come up, but caused them to die a few days later. With the weeds, nearly all that came up were quite certain to survive. The extent of the injury to weeds was shown chiefly by the small number of weeds which appeared on the acid plats as compared with the checks. The failure of seriously injured weed seedlings to appear above ground, as did most of the injured pines, may be due in part to a larger amount of stored food material in the pine seed and in part to a greater depth of soil over many of the weed seeds. It is barely possible that many still dormant weed seeds were killed at the time of the application of the acid. Some of the weed seeds in late-sown plats commence germination at or before the time of acid application, and are therefore probably killed at the time of application. The frequent occurrence of healthy Equisetum in beds where the acid killed the pines may be due entirely to the presence of old rootstocks and not to superior tolerance of acid. It has been suggested that the survival of grass where acid prevented the appearance of dicotyledons may be due to the branching habit of the grass roots, which makes injury to the tip of the primary radicle of less importance than with the plants which depend largely on a main taproot.

Despite the qualifications in the preceding paragraph it seems quite certain that a great many germinating weed seeds which were dormant at the time of the application of the acid and were deeper in the soil, and therefore exposed to lower concentrations of acid than the pines, were killed in much the same way as the pines by amounts of acid which would not injure the pines. The experiments indicate not only a distinctly greater tolerance for sulphuric acid in the pines than in the angiosperms most commonly represented in the beds, but within the angiosperms a somewhat smaller difference in tolerance between the grasses and dicotyledonous species was observed. Tests in water culture would be necessary to establish the differences in resistance of the various species observed in these experiments and to give the differences a quantitative value.

Treatments several days or weeks before sowing also had considerable effect on the number of weeds found in the seed beds during the first few weeks after the germination of the pines. The use of 0.3 ounce of acid 14 days before sowing, with sufficiently frequent watering after sowing to prevent injury to yellow pine, prevented the appearance of any dicotyledons for at least 43 days after treatment and allowed only a few grass seedlings near the edge of the plat and a couple of Equisetum plants. Mollugo, grass, and Portulaca seedlings were common in all the check plats in this series, and Amaranthus and Euphorbia were present, while Equisetum was at least no more common in the checks than in the acid plats.

In another series, in which watering was frequent enough to prevent injury to most pine seedlings, 0.25 ounce of acid nine days before sowing kept the plat free from all weeds except three grass plants for 1½ months, and 0.375 ounce applied at the same time prevented weed growth of any sort. While grasses predominated in the untreated plats, they also contained many plants of Mollugo, Portulaca, Amaranthus, and Euphorbia, their frequency being in the order named.

In another series watered in the same way, 0.281 ounce of acid 11 days before sowing and heavier treatments applied to three other plats at the same time entirely prevented weed growth till 47 days afterwards, while the checks contained the same species as those in the former series.

In series 519, plats A, C, and D (Table VI), 0.25 ounce of acid had a distinct effect on the weed flora, practically the same as 0.375 ounce, in plats examined 66 days after application.

In another series, watered quite frequently after sowing in order to prevent acid injury, acid applied 14 days before sowing the pines was tested. On adjacent plats the upper 6 inches of soil was partially sterilized at about the same time by heating in a moist condition to above 80° C. in an oven, all parts of the soil being brought to at least that temperature and kept there for not less than 10 minutes. The results are presented in Table IV.

TABLE IV.—*Weeds which appeared in plats disinfected by heat and by acid.*

Plat.	Treatment (ounces of acid per square foot).	Weeds found 42 days after treatment.
Four checks	None	60 to 100 per plat; grass commonest, Mollugo and Portulaca frequent, Amaranthus occasional.
J and K	Heated	Grass much as in checks, and making more vigorous growth; 2 or 3 Portulaca plants, and 1 Amaranthus in each plat.
C	0.25	5 grass seedlings, with several Mollugo near edge.
F	.375	5 grass, with 1 Portulaca and 1 Mollugo near edge.
G	.375	4 grass.
D	.5	3 grass.

Evidently, unless the grass seed survived a temperature of 80° C. or more, it had been blown into plats J and K after treatment, and migratory ability may explain part of its predominance over the dicotyledons in acid-treated plats. The results in general, nevertheless, indicate that it is somewhat more resistant to acid than the dicotyledons.

RELATION BETWEEN TIME OF APPLICATION AND AMOUNT OF INJURY.

The foregoing experience with pines and other plants in beds treated with acid at the time of germination, at sowing time, and at various times before sowing, shows clearly, as would be expected, that

the time of germination is when acid applied to the beds will do the most damage to pine seedlings. The longer the period before or after germination takes place that the acid is applied the less danger there is of acid injury. The free acid in the soil solution would normally be decreased by diffusion or leaching downward into the subsoil, by adsorption or solid solution by the soil, and by chemical interaction with other constituents of the soil or soil solution. No attempt has been made to determine the relative importance of these different processes in the removal of the acid from the solution. It has seemed rather surprising that even with applications of acid as small as 0.25 ounce per square foot enough acid remains free in the surface soil three weeks after application to kill the tips of jack-pine roots and prevent the growth of most dicotyledonous weed species for 1½ months. In soil containing large quantities of carbonates there could be no such length of persistence of free acid.

The amount of injury occurring in plats treated at different lengths of time before germination and the comparative lack of relationship between the amount of acid used and the extent of injury in cases where more than 15 days elapse between treatment and germination indicate that the rate of dissipation of the free acid in the soil solution decreases rapidly as the concentration decreases. Very small amounts of acid have proved extremely injurious to root tips in the soil at the time of application. While they lose this extremely toxic character in a very few days after application, the final reduction to a point where no injury occurs requires a relatively long time. The apparent relative stability of very low concentrations of acid in the soil solution is in agreement with the general course of removal of a solute either by diffusion or chemical reaction.

ADDITION OF NEUTRALIZING AGENTS AFTER THE APPLICATION OF THE ACID.

In different experimental series, plats treated with sulphuric acid before sowing were later treated with neutralizing agents to prevent acid injury. This procedure greatly decreased the effectiveness of the acid treatment against the damping-off parasites on whose account the work was being conducted, and so it was not exhaustively tested. In no case was lime applied to the extent of equivalent weights of the acid used.

The indications are that injury to pines may be prevented by small amounts of lime put on the beds a few days after the application of the acid. The results of the treatments are given in Table V.

TABLE V.—*Injury to roots in plats treated with sulphuric acid 12 to 16 days before sowing and later treated with lime.*

Plat.	Treatment (per square foot). Fluid ounce of acid.	Pints of solution.	Avoirdupois ounce of $CaCO_3$.	Days from acid treatment to— Lime application.	Germination.	Excess of acid over lime (fluid ounce per square foot).[1]	Percentage of acid which lime should neutralize.[1]	Injury to pines.	Weeds present 1½ months after acid application.
Pinus ponderosa (series 504-E)	0.375	2	0.250	14	25	0.240	36	None.	Half as many as in check plats of same series.
P. divaricata (series 507): Five checks	None.	....	None.	......	......	......	....	....	Grass and Mollugo abundant; some Amaranthus, Portulaca, and Euphorbia in each plat.
M	.375	3	.250	6	20	.240	36	Slight.	Very few, mainly grass and Mollugo, but growth much more vigorous than in checks.
G	.500	2	.333	8	23	.321	36	None.	
K	.500	4	.333	9	24	.321	36	...do..	
P	.500	4	.200	7	22	.392	22	...do..	
E	.750	4	.333	7	22	.571	24	...do..	
L	.750	2	.500	7	22	.481	36	...do..	
O	.750	6	.500	7	22	.481	36	Slight.	
F	None..	....	.125	......	......	......	....	None..	More weeds than in untreated plats, in vigorous condition.
C	...do...	....	.250	......	......	......	....	...do...	
R	...do...	....	.375	......	......	......	....	...do...	
A	...do...	....	.500	......	......	......	....	...do...	
P. resinosa (series 514): Checks	None.	....	None.	......	......	......	....	....	Grass, Mollugo, Euphorbia, Portulaca, and Amaranthus.
J	.500	2	.333	5	24	.321	36	None.	Less than in checks; grass, Mollugo, and Euphorbia.
K	.500	4	.250	5	24	.365	27	...do..	Less than in checks; grass, Euphorbia, and Amaranthus.
N	None.	....	.500	......	......	......	....	...do..	As for check.

[1] Based on equivalent weights, assuming for the commercial sulphuric acid a maximum specific gravity of 1.84 and a purity of 95 per cent. No allowance is made for impurities in the lime.

It appears that at least in plats M and O the acid applied was not reduced to two-fifths of its original amount during the first six or seven days after application. The injury in plat M, with its acid excess of only 0.24 ounce, when compared with the lack of serious injury in other plats with a greater excess of acid (notably plat E, with an excess 2⅜ times as great), is a further indication of the relative stability of weak acid solutions in the soil.

In the case of series 514, acid injury occurred in a plat treated with a relatively small amount of hydrochloric acid, and it is quite certain that only the lime prevented injury in plats J and K.

Ammonia was also tested, following sulphuric acid. On jack pine, 0.750 fluid ounce of acid applied 21 days before sowing was followed a few days later by 0.469 ounce of the strongest commercial grade of ammonia. No injury to pines occurred. Watering in this series was very frequent, so injury might have taken place with ordinary watering despite the lime used. In series 514, a red-pine plat treated with 0.562 ounce of acid 13 days before sowing, followed by

0.5 ounce of ammonia eight days before sowing, suffered no injury. In this case the heavy acid treatment would probably have resulted in injury had not the ammonia been applied.

From the practical standpoint, the prevention of injury from acid in pine seed beds by the use of neutralizing agents at this nursery is not a success, because beds so treated are often as badly infested by parasites as beds which have received no disinfectant treatment. The action of heavy applications of lime on the beds is also somewhat in question. Amounts up to 0.5 ounce per square foot, as used in the neutralizing work, have, however, been used alone without any bad effect. In one case 0.73 ounce per square foot (equivalent to 1 ton per acre) used on jack-pine beds at or before seeding in two different series was followed by a serious decrease of germination, and in the other case by a marked increase in the number dying after the seedlings came up. Whether the effect was a direct injury to the seedlings or a stimulation of the parasites which attack them was not determined.

The effect on weeds of acid followed by lime is also shown in Table V. Much injury to weeds occurred despite the neutralization several days later of two-fifths of the acid applied. However, it is quite certain, especially in the case of series 504, plat E, that much more injury would have occurred had not the lime been applied. Three-fourths as much acid applied to another plat in this series at about the same time, and not followed by lime, prevented the growth of angiosperms on the plat. The extremely rapid growth of the weeds on the acid-lime plats a few weeks after the application of the lime indicates that most of the remaining acid had been broken down by the lime. If enough lime had been used to neutralize one-half or three-fifths of the acid applied, it is entirely probable that all of the acid remaining at the time of the lime application would have been broken down and the soil rendered entirely safe for sowing any crop plant desired. Because the lime applied was not sufficient to take up at once all the acid remaining in the soil at the time of application, as indicated by the injury to the pines in series 507, plats M and O, the question as to whether the acid prevented weed growth largely by killing dormant seed or entirely by killing germinating seed, as with the pines, remains undecided.

Ammonia, 0.469 ounce per square foot, was used in 3 pints of water a few days after the application of 0.750 ounce of acid, with watering sufficient to prevent injury to jack pine even on unneutralized acid plats. Examination approximately 45 days after the ammonia application showed an entire absence of weeds on the acid-ammonia plat, as on the acid plats, while the four checks all contained plants of grass, Mollugo, Amaranthus, and Portulaca. For 37 days after the ammonia was applied 0.562 ounce of acid followed by 0.5 ounce of ammonia five

days later resulted in preventing most weed growth, but not all. Ammonia alone, 0.5 ounce per square foot, had no effect on the weed stand 65 days after application.

TESTS OF MISCELLANEOUS DISINFECTANTS.

Tests were also made with disinfectants other than sulphuric acid. These are summarized in Table VI, together with enough sulphuric-acid tests to afford a basis for comparison. Because a plat can be directly compared only with the others sown at the same time, the plats are grouped by series rather than by disinfectants.

TABLE VI.—*Injury to pines and weeds by miscellaneous disinfectants.*

Plat.	Disinfectant.			Days from treatment to—		Injury to pines.	Weeds present in plats.
	Substance used.	Per square foot.		Sowing.	Weed examination.		
		Ounces.	Solution.				
			Pints.				
Pinus divaricata:[1] 60	Copper carbonate (basic). Ammonia	0.017 .275	1.4	......	......	Germination reduced to less than one-sixth of that in checks. Nearly all seedlings which came up were severely injured.	
62	Sulphuric acid	.172	1.4	0	......	Injury not serious.	
64	....do	.257	1.4	0	......	....do	
P. ponderosa:[1] 415[2]	Formalin	.25	2	0	......	Germination reduced to less than one-quarter that on other plats. No death due to disinfectant after germination.	
416[2]	....do	.125 .125	2 2	29 0	......	None.	
P. divaricata:[1] Series 501 (8 checks).	None	......	......	......	......	......	Portulaca and grass abundant;[3] *Amaranthus retroflexus* common.
J	Hydrochloric acid.	.188	2	0	30–31	None	One-half or two-thirds as many as in checks.
C	Nitric acid	.375	3	0	30–31	Slight or none	Grass rare; Portulaca, 2 or 3 plants.
B	Sulphuric acid	.125	2	0	30–31	Slight	None.
D	....do	.141	1.5	0	30–31	Moderate to heavy.	Do.
H	....do	.188	2	0	30–31	Moderate	A single trifoliate legume.
L	....do	.25	2	0	30–31	Very heavy	None.
P. ponderosa:[4] Series 504 (5 checks).	None	......	......	......	......	......	Mollugo, grass, and Portulaca abundant; Amaranthus and Euphorbia frequent.
L	Formalin[5]	.562	3	14	43	None	Half as many as in checks, mainly grass.
A	Sulphuric acid	.281	2	14	43	....do	Grass, at edge only; Equisetum, 2 plants.
C	....do	.188	3	0	29	....do	Grass, 2 plants.

[1] Watering as in ordinary nursery practice.
[2] Germination exceptionally rapid.
[3] "Abundant" indicates usually 100 or more plants per plat; "frequent" indicates 20 or more; "common" indicates intermediate numbers. All plats measured 2 by 4 feet.
[4] Watering very frequent.
[5] Plats covered tightly for 3 days after treatment to prevent too early evaporation.

TABLE VI.—*Injury to pines and weeds by miscellaneous disinfectants*—Continued.

Plat.	Disinfectant.			Days from treatment to—		Injury to pines.	Weeds present in plats.
	Substance used.	Per square foot.		Sowing.	Weed examination.		
		Ounces.	Solution.				
P. divaricata:[1]			*Pints.*				
Series 508 (7 checks).	None						Grass abundant; Mollugo, Portulaca, Amaranthus, and Euphorbia follow in the order named.
N	Formalin[2]	0.375	3	8			
P	do	.375	2	8			
M	do	.562	4.5	8			Grass, 8 or 10 plants in each formalin plat; other species rare.
A	do	.75	3	14	43–51	None detected.	
D	do	.75	3	10			
G	do	.75	3	6			
H	do	1.00	8	14			
L	do	1.00	4	14			
U	Sulphuric acid	.25	2	9	43–51	Moderate	Grass, 3 plants.
S	do	.375	2	9	43–51	do	None.
Series 512[3] (5 checks).	None						Mollugo, common; grass, Portulaca, Amaranthus, and Euphorbia follow in the order named.
K	Hydrochloric acid.	.25	2	10		None	Grass, a dozen or more plants; Portulaca and Euphorbia still less abundant.
F	do	.375	3	10		Very slight	Grass, 16 plants; Portulaca, 2; Euphorbia, 2.
D	do	.562	3	10	47–48	Slight to moderate.	Grass, 4 plants; Portulaca, 2; Mollugo, 1.
A	do	.75	4	10		do	Grass, 6 plants; Portulaca, 2.
C	Nitric acid	.75	3	10		Moderate	Grass, 4 plants; unknown, 1.
G	do	1.00	4	10		Very slight	Grass, 3 plants.
N	Sulphuric acid.[4]	.281	3	11		Very heavy	None.
P	do	.375	4	11		Heavy	Do.
M	do	.688	5.5	11		Moderate	Do.
J	do	.688	2	11		do	Do.
Series 514[5] (5 checks.	None						Grass abundant, followed by Mollugo, Euphorbia, Portulaca, and Amaranthus in the order named.
F	Hydrochloric acid.	.75	2	13	42	None	Grass, 14 plants; Mollugo, 3 or 4.
G	do	.75	4	13	42	Slight	Grass, 11 plants; Mollugo and Euphorbia, several plants around edge of plat.
A	Sulphuric acid	.125 .125	1 1	13 0	42	Heavy; ⅓ affected.	None.
C	do	.188 .188	2 2	13 0	42	Heavy; ½ affected.	Do.
D	do	.25 .125	2 2	13 0	42	do	Do.

[1] Watered daily, 0.3 to 0.4 inch.
[2] Plats covered tightly for 3 days after treatment to prevent too early evaporation.
[3] Watered daily; dry at surface on date of examination.
[4] This plat became extra dry at time of germination.
[5] Watered daily, 0.15 to 0.3 inch, until 3 days before germination. Surface dry at that time; watering done twice daily thereafter.

TABLE VI.—*Injury to pines and weeds by miscellaneous disinfectants*—Continued.

Plat.	Disinfectant.			Days from treatment to—		Injury to pines.	Weeds present in plats.
	Substance used.	Per square foot.					
		Ounces.	Solution.	Sowing.	Weed examination.		
P. resinosa—Continued.			*Pints.*				
K	Sulphuric acid / Air-slaked lime.	0.50 / .25	4 /	13 / 8	42	None	Grass, 15 to 20 plants; Euphorbia and Portulaca, 1 or 2 each.
P	Copper acetate / Air-slaked lime.	.281 / .25	3 /	12 / 8	42	Slight or none	Same abundance as in checks, but in much less vigorous condition.
Series 516[1] (2 checks).	None	......	......	......	......	......	Grass abundant, followed by Mollugo, Portulaca, and Euphorbia in the order named.
A	Hydrochloric acid.	.562	3	0	28	([2])	Grass, 4 plants.
C	Nitric acid	.562	3	17	45	None	Grass, 12 plants; Mollugo, nearly same number.
D	Sulphuric acid	.188	2	0	28	do	Grass, 2 plants.
F	do	.281	3	0	28	do	None.
Series 518[3] (6 checks).	None	......	......	......	......	......	Grass abundant, strongly predominating; Mollugo, Portulaca, Amaranthus, and Euphorbia follow in the order named.
J	Formalin[4]	.375	3	17	45	None	Grass, 14 plants; Mollugo, nearly as many; Amaranthus, 2 or 3.
K	do	.562	3	17	45	do	Grass and Mollugo, a dozen plants; Amaranthus, 2.
M	do	1.00	4	17	45	do	Grass, 4 or 5 plants; Mollugo, 4.
C	Mercuric chlorid.	.063	2	17	45	do	Grass frequent.
D	do / Sodium chlorid.	.063 / .188	2	17	45	do	Grass, 5 or 6 plants; Mollugo, 1; Amaranthus, 1.
A	Mercuric chlorid. / Air-slaked lime.	.094 / .094	3 / 0	17 / 13	45	do	Nearly as many as in nearest checks, but larger proportion of grass than in check.
N	Zinc chlorid	.281	3	17	45	do	Grass, 4 plants; Mollugo, 1.
G	Copper sulphate.	.188	3	17	45	Slight, or none	Grass, 9 plants; Mollugo, 2.
P	Hydrochloric acid.	.375	2	0	28	None	Grass, 10 or 12 plants.
F	Nitric acid	.562	3	17	45	do	More grass than in P; Mollugo and Amaranthus also present.
Q	Sulphuric acid	.125	2	0	28	do	Practically the same as in nearest check.

[1] Watered 0.3 inch twice daily.
[2] A little at a margin missed in watering.
[3] Watered 0.3 inch, usually twice daily; surface of plats never allowed to become dry.
[4] Plats covered tightly for 3 days after treatment to prevent too early evaporation.

TABLE VI.—*Injury to pines and weeds by miscellaneous disinfectants*—Continued,

Plat.	Disinfectant.			Days from treatment to—		Injury to pines.	Weeds present in plats.
	Substance used.	Per square foot.		Sowing.	Weed examination.		
		Ounces.	Solution.				
P. divaricata:[1]			*Pints.*				
Series 519 (6 checks).	None						Mollugo very abundant, followed by grass, Amaranthus, Portulaca, and Euphorbia in the order named.
F	Ammonia[2]	0.5	2	34	66	Record lost	As in checks.
U	Mercuric chlorid.	.063	2	0	32	(3)	None.
V	....do Sodium chlorid.	.063 .188	2	0	32	All seed killed	Do.
R	Lime-sulphur	.313	2	0	32	Three-fourths of the seed killed by unknown factor.	Records lost.
S	....do	.75	2	0	32	Germination good.	None.
L	Ferrous sulphate.	.5	2	0	32	Record lost	Nearly as many as in checks.
K	Cupric sulphate.	.281	3	34	66	Moderate to heavy.	Do.
P	Hydrochloric acid.	.562	3	0	32	Very heavy	Grass, very few plants at edge of plat.
M	Nitric acid	1.125	3	34	66	Record lost	Do.
O	....do Sulphuric acid	.188 .125	1 1	34 0	32	Very slight	Do.
C	....do	.25	2	34	66	Record lost	Grass and Mollugo, each 7 or 8 plants; Amaranthus, 1.
D	....do	.25	3	34	66	....do	Grass, 3 plants.
A	....do	.375	3	34	66	....do	Grass, 7 or 8 plants.
H	Heat, 80° C. or greater for not less than 10 minutes.	(4)		Few	32	None	Grass, 7 or 8 plants; Portulaca, 1 or 2.
G	....do	(5)		Few	32	....do	Same as for H.

1 Watered 0.3 inch, twice daily.
2 Plats covered tightly for 3 days after treatment to prevent too early evaporation.
3 Nearly all seed killed; heavy injury to those which germinated.
4 Upper 2½ inches of soil heated.
5 Upper 6 inches of soil heated.

DISCUSSION OF MISCELLANEOUS DISINFECTANTS.

HYDROCHLORIC AND NITRIC ACIDS.

Hydrochloric and nitric acids were used in series 501, plats C and J; 512, plats A, C, D, F, G, and K; 514, plats F and G; 516, plats A and C; 518, plats F and P; and 519, plats M, O, and P (Table VI). Injury by them seems to take place in just the same way as that caused by sulphuric acid, and the injured seedlings presented the same appearance as those injured by sulphuric acid. (See Pl. I, figs. 2 and 3.) Pine seeds were not killed by the amounts used at sowing, but the apices of the radicles in some plats were killed by the acid residue in the surface soil after germination began. Injury may be prevented, as with sulphuric acid, by waterings sufficiently frequent to prevent the concentration of the acid in the surface soil.

Volume for volume, the hydrochloric and nitric acids used did not seem to differ greatly in their effect on the pine seedlings or weeds, the hydrochloric acid appearing rather the more dangerous. The tests offer little opportunity for direct comparison. As with sulphuric acid, pines were less injured than weeds, and the grasses present seemed more resistant than the dicotyledons. The difference in the effect on jack pine, grasses, and *Mollugo verticillata* shows especially well in series 512 and 519, in whose checks Mollugo was the most common weed.

The tests show clearly the low toxicity of these acids in this soil as compared with sulphuric acid, volume for volume. Comparison of plats C and J of series 501 with plats B, D, and H in the same series indicates that sulphuric acid is three or more times as dangerous to both pines and weeds as nitric acid and much more dangerous than hydrochloric acid. In series 512, sulphuric acid seems two or three times as active against the pines as the other two acids, while the disparity in the action on weeds appears still greater. In series 516, results in plats A and D treated at the same time indicate that sulphuric and hydrochloric acids are equally toxic to the weeds when the amount of hydrochloric acid used is three times the amount of sulphuric. In series 518, plats P and Q, 0.375 ounce of hydrochloric acid per square foot appeared considerably more active against weeds than 0.125 ounce of sulphuric acid used on the adjacent plat. Weight for weight, the disparity between the two acids is much less. While the strengths of the acids used were not determined, a statement of the amounts used indicating relative concentrations of ionic hydrogen would have further decreased and might have entirely obliterated the apparent disparity in action between the three acids, as was found by Kahlenberg,[1] True,[2] and Heald [3] in their work with these acids in water culture. For instance, using for comparison sulphuric acid containing 90 per cent H_2SO_4 and making no allowance for impurities, nitric acid containing 60 per cent HNO_3 would contain, volume for volume, but 43 per cent as much ionic hydrogen, and 30 per cent hydrochloric acid but 31 per cent as much, assuming equally complete dissociation in the dilute solutions of the three acids.

TOXIC SALTS.

Copper sulphate, tested only twice, gave rather contradictory results. In series 518, plat G (Table VI), 0.188 ounce per square foot 17 days before sowing caused little or no injury to pines and consid-

[1] Kahlenberg, Louis, and True, R. H. On the toxic action of dissolved salts and their electrolytic dissociation. *In* Bot. Gaz., v. 22, no. 2, p. 81-124, 1896.

[2] True, R. H. The toxic action of a series of acids and of their sodium salts on Lupinus albus. *In* Amer. Jour. Sci., ser. 4, v. 9, no. 51, p. 183-192, 1900.

[3] Heald, F. D. On the toxic effect of dilute solutions of acids and salts upon plants. *In* Bot. Gaz., v. 22, no. 2, 1896, p. 130.

erable injury to weeds, while in series 519, plat K, an amount 50 per cent greater, 34 days before sowing, with more frequent watering, caused considerable injury to pines and had little effect on weeds. Copper sulphate injured pines just as did the acids, by stopping elongation of the radicles shortly after they emerged from the seed. Recovery took place in many cases. A marked case of the production of laterals in recovery from copper-sulphate injury is seen in a seedling taken by Dr. T. C. Merrill from a bed in a similar soil at Garden City, Kans., which had been treated heavily with copper sulphate at sowing and again after germination (fig. 2). Normal yellow pine at this age should have a single straight taproot going down at least five times as far as the one figured and with relatively little development of laterals. Ferrous sulphate (series 519, plat L) gave little evidence of toxic action in the soil as compared with other substances used. Further tests are necessary to give comparable data as to the behavior of copper acetate in the soil and the effect of lime in preventing injury by copper salts, the test made (series 514, plat P) being insufficient. The results in series 518, plat N, indicate that zinc chlorid is as dangerous to weed roots in this soil as copper sulphate, or slightly less dangerous.

Mercuric chlorid in the amounts used acts differently from any of the substances previously mentioned, in that it kills dormant pine seed in the soil at Halsey at the time of application. In series 519, plat V (Table VI), the seeds which failed to germinate were taken out of the soil and carefully examined, both with a hand lens and with a compound microscope. No indication was found that they had ever commenced germination. The difference is presumably due to greater penetrative power. Mercuric chlorid in the soil is injurious both to the roots of seedling pines and to weeds in quantities, which in the case of the other salts tested would have no effect. The addition of common salt to the mercuric chlorid at the time of application appears to increase the damage it does in the soil, possibly by delaying the entire breaking down of the disinfectant until it has time to act on the plants. (Compare 518–C and 519–U with 518–D and 519–V.) The additional toxic effect could hardly have been directly due to the 0.188 ounce of common salt per square foot applied, since 0.2 ounce of salt per square foot applied dry to a jack-pine bed three or four days before sowing in an earlier series had no effect on the pines or on the grass and Mollugo common in the series. The addition of sodium chlorid also makes the disinfectant more convenient to work with by greatly increasing the rapidity of solution. The addition to series 518, plat A, of an amount of air-slaked lime equal in weight to the mercuric chlorid applied four days earlier prevented most of the injury to weeds which occurred with smaller amounts of the chlorid in plats not limed.

With these salts, as with the acids, the pines appeared on the whole more resistant to toxic action than the angiosperms present. There was less evidence in the experiments of a difference in susceptibility to salts in general between the grasses and the dicotyledons. Heald's tests of the resistance of corn and peas to copper salts [1] showed for these plants a reversal of their relative resistance to acid, the peas being able to grow in twice as strong copper solution as corn, whereas with four mineral acids they could grow in solutions only one-fourth as strong.

Ammoniacal copper carbonate was also used with jack pine. A plat of this pine was given a solution made up of 0.006 ounce of copper carbonate and 0.099 fluid ounce of ammonia per square foot the first day after germination, and this was repeated two days later. Eight days after germination the plat was again treated, using 0.014 ounce of carbonate and 0.22 ounce of ammonia per square foot. Practically all the seedlings were killed by these treatments. Most of the injury appeared to be done by the first two applications, in which a total of 0.012 ounce of carbonate per square foot was applied. This plat, which received a total of 0.026 ounce of copper carbonate, was resown 16 days after the last application. No serious injury occurred to the second sowing.

Another plat treated just before sowing (plat 60, Table VI) further indicated a very great toxicity for ammoniacal copper carbonate if only the amount of copper contained is considered. The injury to pine in this plat was much more severe than in plat 64, which had been treated with sulphuric acid more than 25 times the weight of the copper carbonate used on plat 60. It is probable that the extremely toxic action of this fungicide was due more to the action of the ammonia than to the copper. The known tendency of ammonia to prevent the precipitation of copper salts from solution may, however, result in more prolonged activity of the copper in this disinfectant than when simple aqueous solutions of copper salts are applied to the soil.

FORMALIN.

Like mercuric chlorid, formalin is capable of killing seed outright if applied at the time of sowing. In a test of yellow pine in which the disinfectant was applied at sowing (plat 415, Table VI) most of the seeds were killed before they gave any outward evidence of commencing to germinate. So far as could be learned, those which were able to start germination were uninjured. In plat 416 (Table VI), which received the same amount of formalin, half at the time of sowing and half at an interval of a month earlier, no injury could be detected. In all other cases, formalin was applied several days

[1] Heald, F. D. Op. cit., p. 152.

before sowing and did no perceptible damage to pines or pine seed. This was true even in series 508, plat G, in which 0.75 fluid ounce per square foot was applied six days before sowing and evaporation allowed for only three days before sowing. The effect of formalin on the weed stand seemed approximately equivalent to that obtained with one-half or one-third the volume of sulphuric acid. As the weight of H_2SO_4 per fluid ounce of acid used was at least four times as great as the weight of HCHO in the formalin, the formaldehyde appears rather more effective, weight for weight, in keeping down weeds. The radical difference in the type of action of the formalin against the pines renders impossible any direct comparison with acid.

LIME-SULPHUR SOLUTION.

The results in series 519, plats R and S (Table VI), are contradictory. Injury to pines from fairly heavy applications of lime-sulphur at the time of sowing can probably be prevented by sufficient watering during the germination period. The injury to weeds occurred despite heavy watering.

EXPERIMENTS AT MORRISVILLE, PA.

During the season of 1912, in pursuance of recommendations by the writer, tests with sulphuric acid were conducted by Mr. R. E. Lee, under the direction of Mr. John Foley, forester of the Pennsylvania Railroad, at the nursery near Morrisville, Pa. Sulphuric acid only was used. All treatments tested resulted in a decreased stand. The results of very weak treatments on beds given ordinary nursery watering are shown in Table VII.

TABLE VII.—*Evidence of injury to pines by sulphuric acid applied at the time of sowing, Morrisville, Pa.*

Plat.	Number of plats averaged.	Species.	Sowing to germination.	Acid per square foot.	Watering.	Final stand, taking average of checks as 100.	Decrease in stand apparently due to acid.
			Days.	*Fluid oz.*			*Per cent.*
Series 631	1	Pinus ponderosa	6 to 9	0.031	Only as in ordinary nursery practice.	91	9
	11	do	6 to 9	.042		95	5
	8	do	6 to 9	.083		90	10
Series 632	16	Pinus resinosa	9 to 10	.083		55	45
Series 633	21	Pinus strobus	16 to 21	.083		27	73
Series 634	7	Pinus sylvestris	7	.083		74	26
Series 635	17	do	9 to 12	.083		63	37
Series 636	7	Picea excelsa	11	.083		56	44
Series 637	14	Pinus sylvestris	8 to 10	.083		62	38
Series 638:							
A	1	do		.188		34	66
D	1	do		.25		26	74
C	1	do		.375		4	96
G	1	do		.188	0.15 inch, twice daily.	83	17
F	1	do		.25		53	47
J	1	do		.375		45	55

The relative resistance of *Pinus ponderosa* to the acid is probably due to its rapid growth, as at Halsey. The severe injury to *P. strobus* is rather surprising in view of the length of time which elapsed before germination. The consistent relation in series 638 between the decrease in stand and the amount of acid used and the evidently helpful effect of frequent watering leave no reasonable doubt as to the agency of the acid in causing the decreased stand. In all of the series except 631 the treated plats were uniformly poorer than the checks. In series 638, fewer seedlings appeared in acid plats than in the checks in all cases, the deficiency being greatest in the ordinary watering plats, and the amount of death just after the seedlings came up in the ordinary watering plats was very large. The amount of germination and early loss for the other series was not determined.

The evidence of the experiments at Morrisville as a whole shows that at this nursery the amounts of acid necessary to cause injury were much smaller than at Halsey.

GENERAL DISCUSSION AND CONCLUSION.

It is evident that the toxicity of disinfectants to the roots of plants in soil at Halsey, Nebr., varies greatly in response to a number of different factors. The amounts of water in different parts of the soil at different times and the movements of soil water, which result in concentrating the soil solution at particular points, must be considered, as well as the concentration of the solution applied. The depth of the root tips in the soil at the time of greatest concentration of the soil solution is also of prime importance, and the time of application is a very important variable.

In general, while it is evident that disinfectants do not act on plant roots in soil to the same extent as in liquid cultures, they seem to act in much the same way. If only the free poison in the soil solution is considered, it is doubtful whether a great difference in degree of toxicity can be found in soil and in liquid cultures. However, the activity of poisons in the soil solution should not be expected to equal their activity in pure water cultures. Antitoxic relations which have been found by various workers to exist between numerous substances in water cultures may be expected to exist between most disinfectants and various components of the soil solution. An investigation of antagonism between substances obtained in soil extracts and some of the substances used in soil disinfection should yield some interesting results. Most poisons are of necessity rather unstable substances, and even where leaching is prevented, as in pot experiments, and nonvolatile substances are used, the loss of free poisons from the soil solution by combination with soil constituents and by other absorptive processes is undoubtedly great.

That acid solutions, in fact, are much more toxic just after application is clearly shown by the experiments at Halsey. That the rapidity with which disinfectants are rendered inactive in the soil should vary greatly in different soils is to be expected, in view of the great differences which exist in both their physical and chemical constitution. However, examinations of soils by the usual methods of chemical and physical analyses and lime requirement and wilting-coefficient determination do not give much indication as to how sulphuric acid may be expected to behave in different soils. A soil with a low wilting coefficient may be expected to have a rather low average water content under field conditions and therefore to require small amounts of disinfectants to raise the soil solution to a killing concentration. Coarse texture indicates low absorptive power and a consequent small capacity for disinfectants without injury to roots. A high lime requirement may indicate soil acidity, but it may also be found in a nonacid soil which has high absorptive capacity.[1] Both on theoretical grounds and from the results obtained at Halsey with sulphuric acid and mercuric chlorid treatments followed by lime, the carbonates present should have a decided influence in preventing injury by acids, and probably by many toxic salts as well. Experiments are under way at several nurseries at which preliminary results indicated a distinct relation between determinable chemical and physical characters and the behavior of disinfectants. But between soils as much alike as those on which the foregoing experiments were conducted, the physical and chemical examination made gives no clue to any difference which can explain the different behavior of sulphuric acid on the two. Neither soil yielded CO_2 by the method employed in the examination by the Bureau of Soils. The surface soil at Morrisville contains more CaO, has a higher ignition loss, a higher wilting coefficient, and a lower lime requirement than the Halsey soil, all of which would seem to indicate a greater capacity for acid at Morrisville. The experiments show throughout that the reverse is the case. While the tests made at the two nurseries are not absolutely comparable, comparison of the plats of series 501 (Table VI), which received from 0.125 to 0.25 ounce per square foot, with series 631 to 637 inclusive (Table VII), in which from 0.031 to 0.083 ounce was used, indicates that at Halsey the amount of acid required to cause injury is three times that required at Morrisville. It seems probable, in view of the semiarid conditions at Halsey, that the Morrisville soil was more acid or less alkaline than the Halsey soil.

An attempt to get an indication of different reaction between the two soils a year after the samples were taken failed, both soils giving

[1] Cameron, F. K. The Soil Solution, the Nutrient Medium for Plant Growth, p. 65, footnote 1. Easton, Pa., 1911.

negative results with the potassium nitrate and iodin test outlined by Loew [1] and turning blue litmus red, the latter phenomenon likely indicating absorption rather than acid reaction for either soil.[2] Titration of extracts from fresh samples of these soils should give more indication of the real cause of the different behavior of acid at the two places. If difference in reaction of the two soils explains the different results, it is probable that the difference in capacity for disinfectants would be less marked or even reversed with such disinfectants as copper sulphate.

Further evidence of the failure of chemical analysis or physical characters to show what action disinfectants will have on roots in different soils is seen in the difference between the results in these nursery soils and the results obtained by Lipman and Wilson [3] with a soil described as sandy and having a chemical constitution showing no very radical differences from those reported in the foregoing. On this soil they found that there was no evidence of damage to either wheat or vetch seedlings by sulphuric acid in the amount of 600 parts per million of water-free soil applied several days before sowing. While these experiments, conducted in pots, can not be directly compared with those of the writer, it is sufficiently evident that the results are very different. At Halsey, 0.125 fluid ounce per square foot, followed by the ordinary watering given germinating seed beds, entirely prevented the growth for at least a month after application of the monocotyledonous and dicotyledonous weeds represented in the seed beds. Assigning to the commercial acid used a maximum strength, which may be assumed as having a specific gravity of 1.84 and purity of 95 per cent, and to the soil a minimum weight, which for this fine sand may be taken as 80 pounds per cubic foot, we find that even if all the acid applied were held in the upper 4 inches of soil the weight of H_2SO_4 used was only 534 parts per million of soil. That this treatment should have prevented all growth of weeds, in which both monocotyledons and dicotyledons were represented, while 600 parts did not even decrease the growth rate of wheat and vetch on the soil used by Lipman and Wilson, indicates a very considerable difference in behavior of acid in the two soils. As injury to pines is caused on the Morrisville soil by amounts of acid only one-third of that required to injure pines at Halsey, the contrast in the results between the Morrisville soil and that used by Lipman and Wilson is still more marked.

The observations made by the writer on the species of Equisetum, pines, grasses, and dicotyledons most common in the seed beds at

[1] Loew, Oscar. Studies on acid soils of Porto Rico. Porto Rico Agr. Exp. Sta. Bul. 13, p. 6, 1913.

[2] Cameron, F. K. Op. cit., p. 66.

[3] Lipman, C. B., and Wilson, F. H. Toxic inorganic salts and acids as affecting plant growth. *In* Bot. Gaz., v. 55, no. 6, p. 409–420, 1913.

Halsey indicated a considerable variation in resistance to sulphuric acid between species of these four phylogenetic groups, exceeding the variation between different species in the same group. It further appeared that, for the four main groups represented, the higher the group in the evolutionary scale the greater the susceptibility of its representatives to injury, not only by sulphuric acid but by hydrochloric and nitric acids and by some of the toxic salts. It is understood, of course, that these differences would not be expected to obtain with all species of these groups, and parallel water-culture tests with the species observed by the writer would probably show that some of the differences in susceptibility indicated in the nursery tests were due to other factors than variable protoplasmic resistance. The experiments reported in the foregoing were devised primarily for developing disease-control methods, and interpretation of many of the direct effects on the seedlings is of necessity difficult.

From the practical standpoint, it seems probable that sulphuric acid can not be used alone as a disinfectant for sandy soil soon to be sown with truck crops. This is at least true if the plants to be grown prove as susceptible to acid injury as the dicotyledonous weeds encountered in these experiments seemed to be. However, acid can probably be applied with safety on most soils several days before sowing if air-slaked lime sufficient to counteract three-fifths or more of the acid used is raked into the surface soil just before seed sowing. Sulphuric acid is so much cheaper than formalin that if subsequent lime neutralization is found practicable this acid may in many cases supplant both heat and formaldehyde as a soil disinfectant for work in which immediate reinfection with parasites is not feared. The writer's experience indicates that, aside from the destruction of parasites, soil treatment with acid followed by lime results in a considerable increase in the growth of many plants, in some cases being more prompt and marked than that following heat disinfection.

SUMMARY.

Sulphuric, hydrochloric, and nitric acids, and copper sulphate used in disinfection of seed-bed soil caused injury to the roots of pine seedlings and prevented the development of many species of angiospermous weeds. All cause injury to pines by killing the growing apex of the radicle immediately after the seed germinates. They can be used to disinfect pine seed beds only if the operator knows how to recognize and prevent such injury to the pines. Typical healthy and acid-injured seedlings are shown in Plate I, figures 1, 2, and 3, and a method by which injured seedlings can be distinguished from others is described on page 9. Many injured seedlings later resume root growth and recover (Pl. I, fig. 4, and text figs. 1 and 2). Injury is due to the concentration of the disinfectant in the surface soil

consequent on the capillary rise of the soil solution and the evaporation of water from the soil surface. It is found that in a sandy Nebraska soil all injury can be prevented by very frequent watering during the germinating period (pp. 11–12). It can also be prevented in the case of acid applications by adding lime to the soil shortly after treating with the disinfectant (pp. 21–22). The lime method, while undesirable in the case of pines, is probably the only one which will prevent injury to angiospermous seedlings. The acids can be applied to seed beds at the time of sowing without any injury to dormant pine seed. Formaldehyde and mercuric chlorid in sufficient disinfecting strengths must be used several days before seed sowing, as they are able to kill dormant pine seed in the soil. Formaldehyde applied at or before seed sowing never causes the injury to germinating pines that is caused by the acids and salts.

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